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# Reduction of some Heavy Elements of Agricultural Drainage Water using the Azolla Plant (Azolla)

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The study was conducted at the Faculty of Science, University of Tikrit and included alaboratory experiments to estimate the proportion of some heavy elements (lead, potassium, cadmium, magnesium, iron) in the biomass ofAzolla pinnata plant to estimate its biocontrol ability to treat agricultural drainage water as samples were taken from the Azolla pinnata cultivated in two different types of agricultural drainage water and Tape water to estimate the percentage of elements before treatment in the water and after it at three periods (21,14,7) days. The results revealed Increasing the concentration of potassium, lead and magnesium elements in the extracted biomass of Azolla pinnata samples after treatmentof agricultural drainage water (1,2) while this increase was slight in tap water, as for cadmium, the results showed a decrease in its concentration in extracted biomass of A. pinnatasamples grown in agricultural drainage water (1,2) and tap water, and that the were non-significant differences ($P < 0.05$) in the concentration ofiron element in extracted plant samples grown in agricultural drainage water1 as it remained at its concentration ($2.69 \pm 0.00$) during the three weeks, it also maintained its concentration in the samples of the extracted plant grown in tap water, reaching($2.86 \pm 0.00$), while the result shows there was a slight decrease in the concentration of Iron inthe extracted plant in treated agricultural drainage water 2, reaching (2.86) in the first week, decreasing to ($2.69 \pm 0.00$) in the second and third week.

**Keywords:** Azolla Pinnata, heavy Elements, agricultural wastewater, heavy elements, Tape water, water treatment.

## INTRODUCTION

Azolla plant (A. pinnata) is an aquatic plant belonging to the fern family, this plant consists of small plates floating on the surface of the water and forming a dense layer of green plants, azolla usually lives in calm waters, swamps, and lakes with a warm climate (Qiu and Yu, 2003). Azolla is very useful in agriculture and the environment, as it is used to improve soil quality and increase agricultural productivity, as it is used as feed for animals and fish, and as a source of organic fertilizers, in addition to that, Azolla has antistatic propertiesfor the growth of weeds and algae, which makes it used as a natural means of weed control (Volesky, 2019).

Some types of azolla are famous for their ability to fix nitrogen in the soil, through their cooperation with the bacteria in their roots, and this property makes azolla very important in improving soil quality, especially in nitrogen-poor lands, and azolla can be used in other areas as well, such as the manufacture of paper, fibers and medicines, as it is used in scientific research and agricultural experiments due to its flexibility and ease of cultivation and reproduction and is considered one of the sustainable plants, as it is characterized by its rapid reproduction, which means that it can be grown in large quantities without the need to use many natural resources, Azolla also contains a variety of beneficial compounds to the human body such as protein, amino acids, fatty acids, vitamins and minerals, so Azolla is an important source ofPlant nutrition (Cruz, 2020). Azolla is characterized by growing significantly in humid and warm areas in the world, which makes it present in many countries including Africa, Asia, South America, Egypt, Iraq and others, and Azola can be used in the field of aquatic plants that treat water, the term phytotherapy comes from the Greek words python, which means plant and rhidium, which means treatment, This technique is widely used because it is less destructive to the soil than other treatments and the price is generally reasonable, another benefit is the addition of aesthetics to degraded soils and the cultivation of plants. Where plants are used to remove organic and inorganic pollutants from contaminated sites in the water, where these plants play an important role for their ability to grow in freshwater and facilitate biological treatment and water

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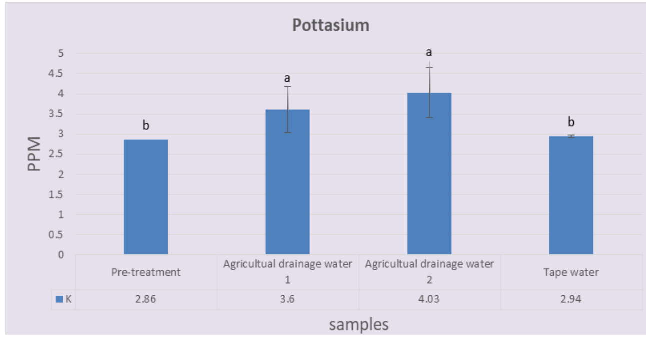
purification from pollutants that cause major environmental problems in the rivers of large industrial cities(Association, 2019),as they are used to improve water quality and purify it from harmful substances andpollutants (Zhu, 2021).

## MATERIALS AND METHODS

**Samples:**Two different samples of agricultural drainage water were collectedfrom different areas of Al-Bu Ajil and Al-Alam in Tikrit district, and the last sample was Tape water.
***Expermental Tests*****:** The concentrations of heavy elements (lead, iron, cadmium, potassium, magnesium) were measured in the Department of Chemical Engineering, Faculty of Engineering, Tikrit University, according to the method followed (APHA,1998) using the atomic absorption spectrophotometer, where 5 grams of extracted A. pinnatasamples were taken and placed in a ceramic container, then the container was transferred to the incineration furnace at a temperature of 600 ° C until the weight stabilized and a white or gray ash was obtained.
The concentration of minerals to be detected were estimated by adding 5 ml of nitric acid of concentration of 5% to the ash obtained and mixed well and then filtered using filter paper and the percentage was estimated using the atomic absorption device.

## RESULTS AND DISCUSSION

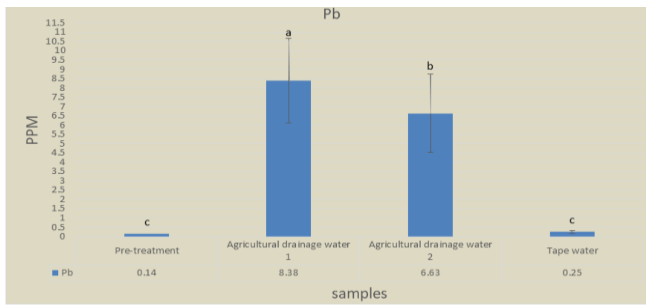
The results of the current study showed a significant increase in the concentration of potassium element (P<0.05) in the biomass of *A. pinnata* plant after 6 weeks of its use in Phytoremediationof agricultural drainage water 1 and 2 compared to the concentrations of potassium in biomass before treatment and biomass after tap water treatment, as Figure (1) and Table(1) show an increase in potassium concentration in biomass after 6 weeks of agricultural wastewater treatment 1 and 2, and reached (3.6±0.57) and (4.03±0.62) ppm respectively compared to (2.8 6) ppm for biomass concentration before treatment, and (2.94±0.03) ppm in biomass after tap water treatment.
This efficiency was lower when treating tap water, as Figure (1) and Table (1) show that there were no significant differences (P<0.05) in the concentration of potassium in the biomass of the plant compared to its concentration before treatment, as it reached (2.94±0.03) ppm after 6 weeks of treatment compared to (2.86) ppm before treatment.




*Different letters indicate significant differences between different water types at a P≤0.05 probability level*

**Figure 1. Bioaccumulation of Potassium element (PPM) in Azolla pinnata plant after treatment.**

Figure (2) and Table(1) show a significant increase (P<0.05) in the concentration of lead element in the biomass of *A. pinnata* used in the treatment of agricultural wastewater 1 and after 6 weeks compared to its concentration before treatment and reached (8.38±2.28) ppm compared to (0.14) ppm before treating, and the concentrations of lead were significantly high (P<0.05) and clearly in the biomass of *A. pinnata* Used in the phytoremediation of agricultural drainage water 2 after completion of treatment and amounted to (6.63±2.12) PPM compared to (0.14) PPM for lead accumulated in biomass before treatment.




*Different letters indicate significant differences between different water types at a P≤0.05 probability level*

**Figure 2. Bioaccumulation of lead (PPM) in *Azolla* pinnata plant after treatment.**

**Table 1. Mineral concentration in the biomass of A. pinnata extracted after treatmentof three different water samples.**

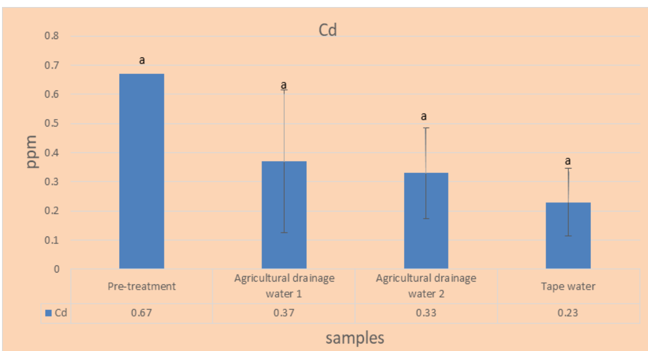
| Minerals (ppm) | Before transplanting | Agricultural wastewater 1 | Agricultural wastewater 2 | Tape water |
|---|---|---|---|---|
| Pb | 0.14c | 8.38±2.28a | 6.63±2.12b | 0.25±0.07C |
| Cd | 0.67a | 0.37±0.24a | 0.33±0.16a | 0.23±0.11A |
| Fe | 2.86a | 2.69±0.00a | 2.69±0.02a | 2.86±0.00A |
| K | 2.86b | 3.60±0.57a | 4.03±0.62a | 2.94±0.03A |
| Mg | 11.31b | 11.90±0.12b | 12.53±0.36a | 11.40±0.01B |

*Different letters in row mean a significant difference at (P≤0.05)*

This increase in the concentration of lead in biomass after agricultural drainage water treatment 1 and 2 was significantly higher compared to its concentrations in plant biomass after treatment of tap water, in which the concentration of lead element was (0.25±0.07) ppm.

The plant did not show any efficiency in reducing lead significantly ($P>0.05$) when used in the treatment of tap water (0.25±0.07) ppm compared to (0.14) ppm before treatment.

Figure (3) and Table (1) show a non-significant decrease ($P>0.05$) in plant efficiency in reducing cadmium in *A. pinnata* biomass after use in the treatment of all types of water samples in the present study, and its concentration were (0.37±0.245, 0.33±0.156 and 0.23±0.115) ppm in *A. pinnata* biomass. And respectively after biotreatment of agricultural drainage water 1, agricultural drainage water 2 and tap water compared to (0.67) ppm for cadmium concentration in biomass before treatment.




*Different letters indicate significant differences between different water types at a P≤0.05 probability level.*

**Figure 3. Cadmium bioaccumulation (PPM) in *Azolla pinnata* after treatment.**

The biomass of *A. pinnata* extracted after the treatment of the three samples of water samples and after 6 weeks of treatment did not show significant differences of ($P>0.05$) in the concentration of iron compared to (2.86) ppm before treatment, as its concentrations in plant biomass after treatment of agricultural drainage water 1, agricultural wastewater 2 and tap water were (2.69±0.00, 2.60±0.02 and 2.86±0.00) ppm respectively as shown in Figure 4 and Table 1.

Figure 5 and Table 1 shows a significant increase ($P<0.05$) in magnesium concentrations in *A. pinnata*biomass after completion of the biocontrol ofagricultural drainage water 2 only and reached (12.53±0.36) ppm compared to (11.31) ppm before treatment. Also, compared with its concentrations in plant biomass after treatment of agricultural drainage water 1 (11.9±0.12) and tap water (11.4±0.01) ppm which showed no significant differences ($P>0.05$) compared to the pre-treatment control sample.

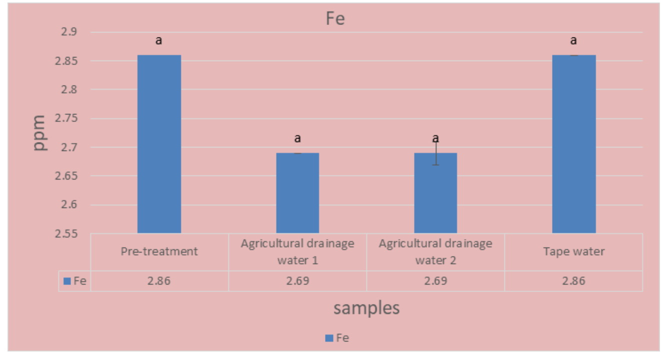



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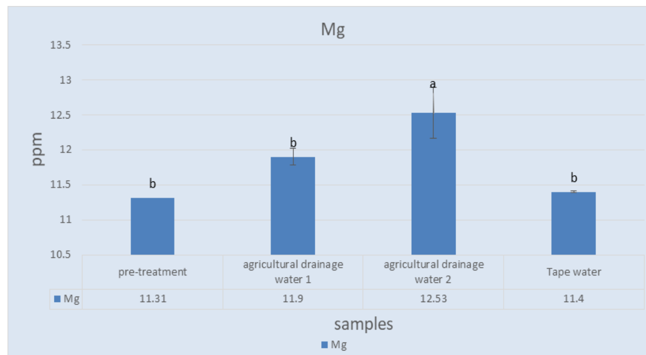
**Figure 4. Bioaccumulation of Iron (PPM) in *Azolla pinnata* plant after treatment.**




*Different letters indicate significant differences between different water types at a P≤0.05 probability level*

**Figure 5. Bioaccumulation of magnesium (PPM) in *Azolla pinnata*biomass.**

Heavy metals pose a serious threat to the environment due to three main criteria: persistence, bioaccumulation, and toxicity. Heavy metals are non-biodegradable chemical residues that possess long persistence in the environment and gradually enter food chains and accumulate at higher trophic levels, endangering animal, and human life. Therefore, reducing or diluting heavy metals from polluted aquatic environments is of great importance in protecting both the environment and human health (Mandakini *et al*., 2016), so the current study aimed to use an environmentally friendly method to dispose of a group of metals in different water samples.

*A. pinnata*has a high bioaccumulation capacity for minerals from polluted water bodies (Wagner, 1997). Numerous ex situ studies have been conducted, including (Salt et al., 1995; Jangwattan, 2010; Sood *et al*., 2011; Moradi *et al*., 2013; Thayaparan *et al*., 2013) all showed the large ability of *A. pinnata* to take and retain various minerals, and all these studies agree with the results obtained in the present study. It can be concluded that *A. pinnata* can be used to dispose of heavy metals in contaminated water bodies.

The basic characteristics of large aquatic plants with strong plant processing capabilities are theirrapid growth rates, higher biomass, and greater adaptability to a wide range of environmental conditions. Moreover, the free-floating nature of*A. pinnata* and other large aquatic plants facilitates harvesting and their high water content in fresh biomass (90-94%), which significantly reduces the volume after drying, and greatly solves the dilemma of disposing of harvested material. A. pinnata dry biomass can easily be transported to recycling sites and used for plant treatment of contaminated water (Sood, 2012). Furthermore, *A. pinnata*has the ability to survive and endure highly polluted water with varying degrees of pH, temperature and salinity, reflecting its suitability for phytotherapy applications.

There are many sources of emission of heavy metal ions into the environment, including waterways. CR enters the environment through disposable batteries, anodizing and other metal finishing processes, tanning, paint and textile industries while Pb is released from disposable batteries, paints, fossil fuel combustion and runoff from road sediments. Although cadmium is largely produced by triphosphate agrochemicals (TSP) containing between 23.5 to 71.7/kg (Noble *et al*., 2014) cadmium is thought to be the main contributing factor to some fatal diseases.

Heavy metals can be removed from aquatic environments through a variety of techniques that include chemical, physical and biological techniques such as sedimentation, ion exchange, chemical reduction, oxidation, membrane filtration, solvent extraction, reverse osmosis and activated carbon adsorption. However, these applications have many limitations, especially in developing countries because they are not economically viable and require highly sophisticated equipment that can be expensive to obtain.

The reason for the efficiency of *A. pinnata* in the disposal of minerals under study may be attributed to the effectiveness of bioaccumulation, as one of the widely used techniques to treat the polluted environment is phytotherapy, which relies on exploiting the plant's intrinsic mechanisms for bioaccumulation or detoxification of heavy metals from soil or aquatic environments. Plant treatment is a low-cost green technique that has been proven effective in diverse aquatic systems including reservoirs and ponds. Some well-known plants As hyperaccumulators are able to absorb heavy metals that are of no importance for the metabolic processes of the plant. These plants with high bioaccumulation efficacy and possessing the ability to absorb non-essential heavy metals have a promising future for effective phytotherapy. The mechanism of hyperaccumulator relies on the absorption of heavy metal pollutants by plant roots and their concentration in plant tissues or their decomposition into less harmful forms (Mandakini *et al*., 2016).

A study (Mandakini *et al*., 2016) showed the efficiency of A. pinnata plant in absorbing many metals, including chromium, cadmium, lead and nickel, and these results are consistent with the results of the current study. The efficiency of the plant used in the current study on reducing minerals is consistent with the conclusions of a study (Akinbile *et al*., 2016). The efficacy of*A. pinnata* in eliminating the element iron was approved by the study (Ena *et al*., 2007). The results of a study (Akinbile *et al*., 2019) agreed with the results of the present study in the efficiency of *A. pinnata* in reducing potassium, magnesium and lead.

The variation of *A. pinnata's* ability in the current study in the reduction of different elements confirms that the bioaccumulation capacity of this plant varies according to the type of element, and this corresponds to the results of a study (Eribo and Kadiri, 2016), where the study showed a high ability of *A. pinnata* to reduce iron and the least reduction of magnesium.The low removal of magnesium in all water samples was indicative of the limited effectiveness of Azolla fern in the effective handling of the metal in the system used in The current study contrasts sharply with the results (Akinbile and Yusoff, 2012).

***Conclusion:*** The locally Azolla pinnata shows different abilities in removing elements from agricultural drainage water, as it shows high efficiency in removing Lead and does not have the ability to remove postassium and iron

***Authors' Contributions statement*:** All authors' have same roll in paper preparation

***Conflict of interest*:** The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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